Technical Data Sheet

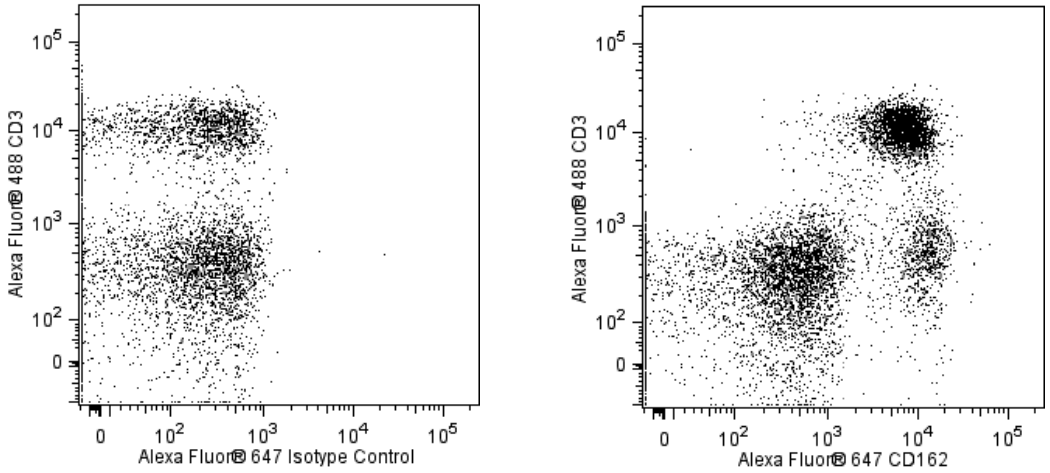
Alexa Fluor® 647 Rat Anti-Mouse CD162

Product Information

Material Number:	**562806**
Alternate Name:	Selplg; PSGL-1; Psgl1; Selp1; Selpl; P-selectin glycoprotein ligand 1
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	2PH1
Immunogen:	Ovalbumin-conjugated peptide covering amino acids 42 to 60 of mouse PSGL-1
Isotype:	Rat (LEW) IgG1, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The 2PH1 monoclonal antibody specifically binds to the N-terminus of CD162 (P-selectin glycoprotein ligand-1, PSGL-1), encoded by the *Selplg* gene. PSGL-1 is expressed on the cell surface as a homodimer of approximately 230 kDa. In the mouse, *Selpl* mRNA is detected in most tissues, with high levels found in hematopoietic cells, brain, and adipose tissue. Flow cytometric analyses have revealed CD162 expression on bone marrow-derived mast and dendritic cells, splenic leukocytes, platelets, peripheral blood neutrophils, and neutrophil and T-cell lines. PSGL-1 is a ligand for P-selectin (CD62P) and is involved in leukocyte rolling, the migration of leukocytes into inflamed tissues, and responses to vascular injury. It is a sialomucin that must be specifically sialylated, fucosylated, and sulfated to bind P-selectin. There is also evidence that other ligands for PSGL-1 and CD62P may exist. The 2PH1 antibody is reported to block binding of mouse leukocytes to CD62P, but the 4RA10 antibody (Cat. No. 557787) has significantly greater blocking activity.



Multicolor flow cytometric analysis of CD162 expression on BALB/c mouse splenocytes. *Splenocytes were stained simultaneously with Alexa Fluor® 488 Rat Anti-Mouse CD3 antibody (Cat. No. 557666) and with either Alexa Fluor® 647 Rat IgG1, κ Isotype Control (Cat. No. 557731; Left Panel) or Alexa Fluor® 647 Rat Anti-Mouse CD162 (Cat. No. 562806; Right Panel). Two-color flow cytometric dot plots show the correlated expression patterns of CD162 (or Ig isotype control staining) versus CD3 for gated events with the forward and side light-scatter characteristics of viable spleen cells. Flow cytometry was performed using a BD™ LSR II Flow Cytometer System.*

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed.

Application Notes

Application

Flow cytometry	Routinely Tested

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Suggested Companion Products

Catalog Number	Name	Size	Clone
557731	Alexa Fluor® 647 Rat IgG1, κ Isotype Control	0.1 mg	R3-34
554656	Stain Buffer (FBS)	500 ml	(none)
557666	Alexa Fluor® 488 Hamster Anti-Mouse CD3e	0.1 mg	145-2C11

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
4. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
5. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
6. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
7. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
8. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.

References

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Borges E, Tietz W, Steegmaier M, et al. P-selectin glycoprotein ligand-1 (PSGL-1) on T helper 1 but not on T helper 2 cells binds to P-selectin and supports migration into inflamed skin. *J Exp Med.* 1997; 185(3):573-578. (Biology)

Frenette PS, Denis CV, Weiss L, et al. P-Selectin glycoprotein ligand 1 (PSGL-1) is expressed on platelets and can mediate platelet-endothelial interactions in vivo. *J Exp Med.* 2000; 191(8):1413-1422. (Biology)

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Li F, Wilkins PP, Crawley S, Weinstein J, Cummings RD, McEver RP. Post-translational modifications of recombinant P-selectin glycoprotein ligand-1 required for binding to P- and E-selectin. *J Biol Chem.* 1996; 271(6):3255-3264. (Biology)

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Phillips JW, Barringhaus KG, Sanders JM, et al. Single injection of P-selectin or P-selectin glycoprotein ligand-1 monoclonal antibody blocks neointima formation after arterial injury in apolipoprotein E-deficient mice. *Circulation.* 2003; 107(17):2244-2249. (Biology)

Sperandio M, Smith ML, Forlow SB, et al. P-selectin glycoprotein ligand-1 mediates L-selectin-dependent leukocyte rolling in venules. *J Exp Med.* 2003; 197(10):1355-1363. (Clone-specific: Flow cytometry)

Steegmaier M, Blanks JE, Borges E, Vestweber D. P-selectin glycoprotein ligand-1 mediates rolling of mouse bone marrow-derived mast cells on P-selectin but not efficiently on E-selectin. *Eur J Immunol.* 1997; 27(6):1339-1345. (Biology)

Xia L, Sperandio M, Yago T, et al. P-selectin glycoprotein ligand-1-deficient mice have impaired leukocyte tethering to E-selectin under flow. *J Clin Invest.* 2002; 109(7):939-950. (Clone-specific: Flow cytometry)

Yang J, Galipeau J, Kozak CA, Furie BC, Furie B. Mouse P-selectin glycoprotein ligand-1: molecular cloning, chromosomal localization, and expression of a functional P-selectin receptor. *Blood.* 1996; 87(10):4176-4186. (Biology)

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